

09/369735

=> d his

(FILE 'HOME' ENTERED AT 10:44:36 ON 21 APR 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 10:45:06 ON 21 APR 2005

L1 2266 S BETA(A) GLYCOSIDASE?  
L2 213 S ASSAY? AND L1  
L3 151781 S OLIGOSACCHARIDE? OR HETEROSACCHARIDE?  
L4 11 S L2 AND L3  
L5 7 DUP REM L4 (4 DUPLICATES REMOVED)  
L6 16 S ALKY(A)CHAIN?  
L7 0 S L2 AND L6  
L8 5827 S BETA(A)GLUCOSIDE  
L9 10 S L2 AND L8  
L10 6 DUP REM L9 (4 DUPLICATES REMOVED)  
L11 786835 S HYDROLYSIS  
L12 23 S L2 AND L11  
L13 14 DUP REM L12 (9 DUPLICATES REMOVED)  
E MATSUI I/AU  
L14 651 S E3  
E ISHIKAWA K/AU  
L15 8696 S E3  
E ISHIDA H/AU  
L16 6232 S E3  
E KOSUGI Y/AU  
L17 467 S E3  
L18 15871 S L14 OR L15 OR L16 OR L17  
L19 6 S L1 AND L18  
L20 2 DUP REM L19 (4 DUPLICATES REMOVED)

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(ROSPATENT) added to list of core patent offices covered  
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NEWS 5 FEB 28 BABS - Current-awareness alerts (SDIs) available  
NEWS 6 FEB 28 MEDLINE/LMEDLINE reloaded  
NEWS 7 MAR 02 GBFULL: New full-text patent database on STN  
NEWS 8 MAR 03 REGISTRY/ZREGISTRY - Sequence annotations enhanced  
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NEWS 13 MAR 22 REGISTRY/ZREGISTRY enhanced with experimental property tags  
NEWS 14 APR 04 EPFULL enhanced with additional patent information and new  
fields  
NEWS 15 APR 04 EMBASE - Database reloaded and enhanced  
NEWS 16 APR 18 New CAS Information Use Policies available online  
  
NEWS EXPRESS JANUARY 10 CURRENT WINDOWS VERSION IS V7.01a, CURRENT  
MACINTOSH VERSION IS V6.0c(ENG) AND V6.0Jc(JP),  
AND CURRENT DISCOVER FILE IS DATED 10 JANUARY 2005  
  
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FILE 'LIFESCI' ENTERED AT 10:45:06 ON 21 APR 2005

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=> s beta(A0 glycosidase?

MISSING OPERATOR 'BETA(A0'

The search profile that was entered contains terms or  
nested terms that are not separated by a logical operator.

=> s beta(A) glycosidase?

L1 2266 BETA(A) GLYCOSIDASE?

=> s assay? and l1

L2 213 ASSAY? AND L1

=> s oligosaccharide? or heterosaccharide?

L3 151781 OLIGOSACCHARIDE? OR HETEROSACCHARIDE?

=> s l2 and l3

L4 11 L2 AND L3

=> dup rem l4

PROCESSING COMPLETED FOR L4

L5 7 DUP REM L4 (4 DUPLICATES REMOVED)

=> d 1-7 ibib ab

L5 ANSWER 1 OF 7 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on  
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ACCESSION NUMBER: 2004:882010 SCISEARCH

THE GENUINE ARTICLE: 858XQ

TITLE: Directed evolution of a glycosynthase from Agrobacterium

sp increases its catalytic activity dramatically and expands its substrate repertoire

AUTHOR: Kim Y W; Lee S S; Warren R A J; Withers S G (Reprint)

CORPORATE SOURCE: Univ British Columbia, Dept Chem, 2036 Main Mall, Vancouver, BC V6T 1Z1, Canada (Reprint); Univ British Columbia, Dept Chem, Vancouver, BC V6T 1Z1, Canada; Univ British Columbia, Ctr Excellence Canada, Prot Engr Network, Vancouver, BC V6T 1Z1, Canada; Univ British Columbia, Dept Microbiol & Immunol, Vancouver, BC V6T 1Z1, Canada

COUNTRY OF AUTHOR: Canada

SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (8 OCT 2004) Vol. 279, No. 41, pp. 42787-42793.  
 Publisher: AMER SOC BIOCHEMISTRY MOLECULAR BIOLOGY INC, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814-3996 USA.  
 ISSN: 0021-9258.

DOCUMENT TYPE: Article; Journal

LANGUAGE: English

REFERENCE COUNT: 52

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB The Agrobacterium sp. beta-glucosidase (Abg) is a retaining **beta-glycosidase** and its nucleophile mutants, termed Abg glycosynthases, catalyze the formation of glycosidic bonds using alpha-glycosyl fluorides as donor sugars and various aryl glycosides as acceptor sugars. Two rounds of random mutagenesis were performed on the best glycosynthase to date (AbgE358G), and transformants were screened using an on-plate endocellulase coupled **assay**. Two highly active mutants were obtained, 1D12 (A19T, E358G) and 2F6 (A19T, E358G, Q248R, M407V) in the first and second rounds, respectively. Relative catalytic efficiencies (k(cat)/K-m) of 1: 7: 27 were determined for AbgE358G, 1D12, and 2F6, respectively, using alpha-D-galactopyranosyl fluoride and 4-nitrophenyl beta-D-glucopyranoside as substrates. The 2F6 mutant is not only more efficient but also has an expanded repertoire of acceptable substrates. Analysis of a homology model structure of 2F6 indicated that the A19T and M407V mutations do not interact directly with substrates but exert their effects by changing the conformation of the active site. Much of the improvement associated with the A19T mutation seems to be caused by favorable interactions with the equatorial C2-hydroxyl group of the substrate. The alteration of torsional angles of Glu-411, Trp-412, and Trp-404, which are components of the aglycone (+1) subsite, is an expected consequence of the A19T and M407V mutations based on the homology model structure of 2F6.

L5 ANSWER 2 OF 7 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2004:1000299 HCAPLUS

DOCUMENT NUMBER: 142:112342

TITLE: Association of the macrophage activating factor (MAF) precursor activity with polymorphism in vitamin D-binding protein

AUTHOR(S): Nagasawa, Hideko; Sasaki, Hideyuki; Uto, Yoshihiro; Kubo, Shinichi; Hori, Hitoshi

CORPORATE SOURCE: Department of Biological Science and Technology, Faculty of Engineering, The University of Tokushima, Tokushima, 770-8506, Japan

SOURCE: Anticancer Research (2004), 24(5C), 3361-3366  
 CODEN: ANTRD4; ISSN: 0250-7005

PUBLISHER: International Institute of Anticancer Research

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Background: Serum vitamin D-binding protein (Gc protein or DBP) is a highly expressed polymorphic protein, which is a precursor of the inflammation primed macrophage activating factor, GcMAF, by a cascade of carbohydrate processing reactions. In order to elucidate the relationship

between Gc polymorphism and GcMAF precursor activity, we estimated the phagocytic ability of three homotypes of Gc protein, Gc1F-1F, Gc1S-1S and Gc2-2, through processing of their carbohydrate moiety. Materials and Methods: We performed Gc typing of human serum samples by isoelectric focusing (IEF). Gc protein from human serum was purified by affinity chromatog. with 25-hydroxyvitamin D3-sepharose. A phagocytosis **assay** of Gc proteins, modified using **.beta.-glycosidase** and sialidase, was carried out. Results: The Gc1F-1F phenotype was revealed to possess Gal $\beta$ 1-4GalNAc linkage by the anal. of GcMAF precursor activity using  $\beta$ 1-4 linkage-specific galactosidase from jack bean. The GcMAF precursor activity of the Gc1F-1F phenotype was highest among three Gc homotypes. Conclusion: The Gc polymorphism and carbohydrate diversity of Gc protein are significant for its pleiotropic effects.

REFERENCE COUNT: 31 THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L5 ANSWER 3 OF 7 LIFESCI COPYRIGHT 2005 CSA on STN

ACCESSION NUMBER: 2004:94752 LIFESCI

TITLE: Physical and Kinetic Properties of the Family 3  $\beta$ -Glucosidase from *Aspergillus niger* Which Is Important for Cellulose Breakdown

AUTHOR: Seidle, H.F.; Marten, I.; Shoseyov, O.; Huber, R.E.

CORPORATE SOURCE: Division of Biochemistry, Faculty of Science, University of Calgary, 2500 University Dr. NW, Calgary, Alberta, T2N 1N4, Canada; E-mail: huber@ucalgary.ca

SOURCE: Protein Journal [Protein J.], (20040100) vol. 23, no. 1, p. 11.  
ISSN: 1572-3887.

DOCUMENT TYPE: Journal

FILE SEGMENT: K

LANGUAGE: English

SUMMARY LANGUAGE: English

AB A  $\beta$ -glucosidase (BG sub(s)) purified from *Aspergillus niger* cellulase powder (obtained from Sigma, St. Louis, MO, USA) was characterized. Electrophoresis, size exclusion chromatography, and dynamic light scattering indicated that the enzyme is a dimer of approximately 200 kDa. Five of the seven N-glycosylated **oligosaccharides** attached to BG sub(s) were composed of D-mannoses attached to a  $\beta$ -(1-4)-N-acetylglucosamine- $\beta$ -(1-4)-fucose- $\alpha$ -(1-6)-N-acetylglucosamine core. The other two were similar, but the cores of these did not have the D-fucose. The enzyme is a retaining glycosidase, and it also has a distinct preference for the  $\beta$ -configuration at the reducing end of cellobiose. BG sub(s) is thermostable up to 65 degree C but is sensitive to freezing and thawing. The extinction coefficient of BG sub(s) was found to be 1.8 cm super(-1) mg super(-1). All substrates **assayed** resulted in Eadie-Hofstee plots that were curved at high substrate concentrations. TLC of the reaction products showed that the substrates themselves act as acceptors when present at high concentrations. The transglucosidic activity rate is different from the hydrolytic activity rate and this causes the curvature at high substrate concentrations. The enzyme produces gentiobiose when D-glucose is the acceptor. pH optima of the V sub(max) {h} with pNPGlc, oNPGlc, and cellobiose were between pH 4 and 4.5, and the K sub(m) values decreased at pH values between 3 and 5. Inhibition experiments indicated that the enzyme is specific for glucosyl substrates and suggested that D-gluconolactone is a transition state analog. Studies with cello-**oligosaccharides** and 3,4-dinitrophenyl-cellobiose showed that BG sub(s) is an exo-hydrolase having at least five glucose subsites and that it cleaves from the nonreducing end. The properties of a family 3  $\beta$ -glucosidase (BG sub(3)) sequenced by Dan et al. was also studied and was shown to have very similar properties to those of BG sub(s). Sequence analysis of a portion of BG sub(s) verified that these are the same enzymes.

L5 ANSWER 4 OF 7 EMBASE COPYRIGHT 2005 ELSEVIER INC. ALL RIGHTS RESERVED.  
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ACCESSION NUMBER: 2003081409 EMBASE  
TITLE: Maillard reactions and increased enzyme inactivation during  
**oligosaccharide** synthesis by a hyperthermophilic  
glycosidase.  
AUTHOR: Bruins M.E.; Van Hellemond E.W.; Janssen A.E.M.; Boom R.M.  
CORPORATE SOURCE: M.E. Bruins, Department of Food Technology, Food/Bioprocess  
Engineering Group, Wageningen University, P.O. Box 8129,  
6700 EV, Wageningen, Netherlands.  
Marieke.Bruins@Algemeen.pk.wau.nl  
SOURCE: Biotechnology and Bioengineering, (5 Mar 2003) Vol. 81, No.  
5, pp. 546-552.  
Refs: 21  
ISSN: 0006-3592 CODEN: BIBIAU  
COUNTRY: United States  
DOCUMENT TYPE: Journal; Article  
FILE SEGMENT: 004 Microbiology  
LANGUAGE: English  
SUMMARY LANGUAGE: English  
ENTRY DATE: Entered STN: 20030306  
Last Updated on STN: 20030306

AB The thermostable *Pyrococcus furiosus* **.beta.-glycosidase**  
was used for **oligosaccharide** production from lactose in a  
kinetically controlled reaction. Our experiments showed that higher  
temperatures are beneficial for the absolute as well as relative  
**oligosaccharide** yield. However, at reaction temperatures of  
80°C and higher, the inactivation rate of the enzyme in the  
presence of sugars was increased by a factor of 2 compared to the  
inactivation rate in the absence of sugars. This increased enzyme  
inactivation was caused by the occurrence of Maillard reactions between  
the sugar and the enzyme. The browning of our reaction mixture due to  
Maillard reactions was modeled by a cascade of a zeroth- and first-order  
reaction and related to enzyme inactivation. From these results we  
conclude that modification of only a small number of amino groups already  
gives complete inactivation of the enzyme. .COPYRG. 2003 Wiley  
Periodicals.

L5 ANSWER 5 OF 7 EMBASE COPYRIGHT 2005 ELSEVIER INC. ALL RIGHTS RESERVED.  
on STN DUPLICATE 1

ACCESSION NUMBER: 2001350554 EMBASE  
TITLE: Production of a lactose-free galacto-  
**oligosaccharide** mixture by using selective  
enzymatic oxidation of lactose into lactobionic acid.  
AUTHOR: Splechtna B.; Petzelbauer I.; Baminger U.; Haltrich D.;  
Kulbe K.D.; Nidetzky B.  
CORPORATE SOURCE: B. Nidetzky, Division of Biochemical Engineering, Institute  
of Food Technology, Univ. fur Bodenkultur Wien (BOKU),  
Muthgasse 18, A-1190 Wien, Vienna, Austria.  
nide@edv2.boku.ac.at  
SOURCE: Enzyme and Microbial Technology, (4 Oct 2001) Vol. 29, No.  
6-7, pp. 434-440.  
Refs: 21  
ISSN: 0141-0229 CODEN: EMTED2  
S 0141-0229(01)00412-4  
PUBLISHER IDENT.:  
COUNTRY: United States  
DOCUMENT TYPE: Journal; Article  
FILE SEGMENT: 004 Microbiology  
LANGUAGE: English  
SUMMARY LANGUAGE: English  
ENTRY DATE: Entered STN: 20011018  
Last Updated on STN: 20011018

AB We report a novel and efficient way of producing lactose-derived galacto-**oligosaccharides** (GOS) that do not contain remaining lactose and monosaccharides. The initial sugar mixture was obtained by enzymatic transformation at 70°C of a lactose solution of 270 g/liter using recombinant **beta-glycosidase** from the Archaeon *Sulfolobus solfataricus*. At the optimum reaction time for kinetically controlled transgalactosylation, it contained 46% monosaccharides, 13% lactose and 41% GOS. Lactose was selectively oxidised into lactobionic acid by using fungal cellobiose dehydrogenase which displays a  $\approx$ 100-fold preference for reaction with lactose compared to reaction with GOS. Oxidation of lactose was coupled to reduction of 2,6-dichloro-indophenol which was added in catalytic concentrations. The oxidised redox mediator was regenerated continuously by fungal laccase-catalysed reduction of molecular oxygen into water. Ion exchange chromatographies were employed to remove lactobionic acid, other ions and monosaccharides. The final product contained 97% GOS, 1.2% lactose and 2.1% monosaccharides. The yield accounted for 25% of original lactose. An enzymatic **assay** for lactose has been developed. It is robust and allows sensitive quantification of the analyte in complex sugar mixtures containing large excesses of monosaccharides and GOS. .COPYRGT. 2001 Elsevier Science Inc. All rights reserved.

L5 ANSWER 6 OF 7 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN  
ACCESSION NUMBER: 1999:490462 BIOSIS  
DOCUMENT NUMBER: PREV199900490462  
TITLE: beta-Galactooligosaccharide synthesis with  
beta-galactosidases from *Sulfolobus solfataricus*,  
*Aspergillus oryzae*, and *Escherichia coli*.  
AUTHOR(S): Reuter, Stephan; Nygaard, Anne Rusborg; Zimmermann,  
Wolfgang [Reprint author]  
CORPORATE SOURCE: Biotechnology Laboratory, University of Aalborg,  
Sohnngaardsholmsvej 57, DK-9000, Aalborg, Denmark  
SOURCE: Enzyme and Microbial Technology, (Sept., 1999) Vol. 25, No.  
6, pp. 509-516. print.  
CODEN: EMTED2. ISSN: 0141-0229.  
DOCUMENT TYPE: Article  
LANGUAGE: English  
ENTRY DATE: Entered STN: 16 Nov 1999  
Last Updated on STN: 16 Nov 1999

AB The synthetic potential of the thermostable **beta-glycosidase** from *Sulfolobus solfataricus* was compared with those of the commercially available, thermotolerant beta-galactosidase (beta-D)-galactoside galactohydrolase, EC 3.2.1.23) from *Aspergillus oryzae* and the nonthermostable beta-galactosidase from *Escherichia coli*. To synthesize beta-galactooligosaccharides at different temperatures, the transglycosylation approach with lactose as donor and N-acetyllactosamine as acceptor was chosen as a model reaction. Generally, the *S. solfataricus* enzyme gave the highest yields (48%), followed by the *A. oryzae* (36%) and the *E. coli* beta-galactosidases (32%). The main products obtained with the thermostable **beta-glycosidase** from *S. solfataricus* were beta-D-Gal-(1-6)-D-GlcNAc, beta-D-Gal-(1-4)-D-GlcNAc and several **oligosaccharides**. The enzyme from *A. oryzae* gave a similar product pattern but lower yields. beta-D-Gal-(1-6)-D-GlcNAc was formed as the main disaccharide by the *E. coli* beta-galactosidase, and other products could only be detected in trace amounts.

L5 ANSWER 7 OF 7 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN  
ACCESSION NUMBER: 1985:254867 BIOSIS  
DOCUMENT NUMBER: PREV198579034863; BA79:34863  
TITLE: EFFECT OF CASTANOSPERMINE ON THE STRUCTURE AND SECRETION OF  
GLYCOPROTEIN ENZYMES IN *ASPERGILLUS-FUMIGATUS*.  
AUTHOR(S): ELBEIN A D [Reprint author]; MITCHELL M; MOLYNEUX R J  
CORPORATE SOURCE: DEP BIOCHEM, UNIV TEXAS HEALTH SCI CENT, SAN ANTONIO, TEX

78284, USA  
SOURCE: Journal of Bacteriology, (1984) Vol. 160, No. 1, pp. 67-75.  
CODEN: JOBAAY. ISSN: 0021-9193.  
DOCUMENT TYPE: Article  
FILE SEGMENT: BA  
LANGUAGE: ENGLISH

AB A. fumigatus secretes a number of glycosidases into the culture medium when the cells are grown in a mineral salts medium containing guar flour (a galactomannan) as the C source. At least some of these glycosidases were reported to be glycoproteins having N-linked **oligosaccharides**. The effect of the glycoprotein processing inhibitor, castanospermine, on the structures of the N-linked **oligosaccharides** and on the secretion of various glycosidases was studied. Cells were grown in the presence of various amounts of castanospermine; at different times of growth, samples of the media were removed for the measurement of enzymatic activity. Of the 3 glycosidases **assayed**,  $\beta$ -hexosaminidase was most sensitive to castanospermine; and its activity was depressed 30-40% at 100  $\mu$ g of alkaloid/ml and even more at higher alkaloid concentrations. On the other hand,  $\beta$ -galactosidase activity was hardly diminished at castanospermine levels of up to 1 mg/ml, but significant inhibition was observed at 2 mg/ml.  $\beta$ -Galactosidase was intermediate in sensitivity. Cells were grown in the presence or absence of castanospermine and labeled with [2-3H]mannose, [6-3H]glucosamine, or [1-3H]galactose to label the sugar portion of the glycoproteins. The secreted glycoproteins were digested with pronase to obtain glycopeptides, and these were identified on Bio-Gel P-4 (Bio-Rad Laboratories). The glycopeptides were then digested with endoglucosaminidase H to release the peptide portion of susceptible structures, and the released **oligosaccharides** were reisolated and identified on Bio-Gel P-4. The **oligosaccharides** from control and castanospermine-grown cells were identified by a combination of enzymatic and chemical studies. In control cells, the **oligosaccharide** appeared to be mostly Man8GlcNAc and Man9GlcNAc, whereas in the presence of alkaloid, the major structures were Glc3Man7GlcNAc and Glc3Man8GlcNAc. These data fit previous observations that castanospermine inhibits glucosidase I.

=> s alky(a)chain?  
L6 16 ALKY(A) CHAIN?

=> d his

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FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 10:45:06 ON 21 APR 2005

L1 2266 S BETA(A) GLYCOSIDASE?  
L2 213 S ASSAY? AND L1  
L3 151781 S OLIGOSACCHARIDE? OR HETEROSACCHARIDE?  
L4 11 S L2 AND L3  
L5 7 DUP REM L4 (4 DUPLICATES REMOVED)  
L6 16 S ALKY(A)CHAIN?

=> s 12 and 16  
L7 0 L2 AND L6

=> s beta(a)glucoside  
L8 5827 BETA(A) GLUCOSIDE

=> s 12 and 18  
L9 10 L2 AND L8

=> dup rem 19



PROCESSING COMPLETED FOR L9

L10 6 DUP REM L9 (4 DUPLICATES REMOVED)

=> d 1-6 ibib ab

L10 ANSWER 1 OF 6 EMBASE COPYRIGHT 2005 ELSEVIER INC. ALL RIGHTS RESERVED.  
on STN DUPLICATE 1

ACCESSION NUMBER: 1998072100 EMBASE

TITLE: Substrate specificities of midgut **.beta.-glycosidases** from insects of different orders.

AUTHOR: Ferreira C.; Torres B.B.; Terra W.R.

CORPORATE SOURCE: C. Ferreira, Departamento de Bioquimica, Instituto de Quimica, Universidade de Sao Paulo, C.P. 26077, 05599-970, Sao Paulo, Brazil. clfterra@quim.iq.usp.br

SOURCE: Comparative Biochemistry and Physiology - B Biochemistry and Molecular Biology, (1998) Vol. 119, No. 1, pp. 219-225.  
Refs: 26

ISSN: 0305-0491 CODEN: CBPBB8

PUBLISHER IDENT.: S 0305-0491(97)00310-6

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology  
029 Clinical Biochemistry

LANGUAGE: English

SUMMARY LANGUAGE: English

ENTRY DATE: Entered STN: 19980326

Last Updated on STN: 19980326

AB **.beta.-Glycosidase** activity was determined in midgut homogenates and in electrophoretically-resolved midgut samples from the following insects: Orthoptera, *Abracris flavolineata*; Coleoptera, *Pheropsophus aequinoctialis*, *Tenebrio molitor*, *Pyrearinus termitilluminans*; Hymenoptera, *Scaptotrigona bipunctata*; Diptera, *Rhynchosciara americana*; Lepidoptera, *Erinnyis ello*, *Spodoptera frugiperda*, and *Diatraea saccharalis*. The substrates used in the assays included disaccharides (cellobiose and lactose), synthetic aryl **.beta.-glucosides** (p-nitrophenyl **.beta.-glucoside** and p-nitrophenyl  $\beta$ -galactoside), and plant glycosides (salicin and amygdalin). Orthopterans, coleopterans, and hymenopterans have in their midgut  $\beta$ -galactosidases (active only on  $\beta$ -galactosides) and class 2 (active only on disaccharides) and class 3 (active only on synthetic and plant glycosides)  $\beta$ -glucosidases. Class 1 (active on disaccharides and on synthetic and plant glycosides)  $\beta$ -glucosidases, at least in orthopterans, have putatively different sites for each activity. Dipterans have  $\beta$ -galactosidases and a single class 1  $\beta$ -glucosidase. Lepidopterans have no  $\beta$ -galactosidases and may have a single class 1  $\beta$ -glucosidase or class 1 and class 3  $\beta$ -glucosidases. There seems to be an evolutionary trend from multiple enzymes with different substrate specificities to a single enzyme able to hydrolyse all the  $\beta$ -glycosides in the same site.

L10 ANSWER 2 OF 6 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1998:318393 BIOSIS

DOCUMENT NUMBER: PREV199800318393

TITLE: The *arbZ* gene from *Lactobacillus delbrueckii* subsp. *lactis* confers to *Escherichia coli* the ability to utilize the **.beta.-glucoside** arbutin.

AUTHOR(S): Weber, Beate A. [Reprint author]; Klein, Juergen R.; Henrich, Bernhard

CORPORATE SOURCE: Fachbereich Biol., Abt. Mikrobiol., Univ. Kaiserslautern, Postfach 3049, D-67653 Kaiserslautern, Germany

SOURCE: Gene (Amsterdam), (June 8, 1998) Vol. 212, No. 2, pp. 203-211. print.

CODEN: GENED6. ISSN: 0378-1119.

DOCUMENT TYPE: Article  
LANGUAGE: English  
OTHER SOURCE: EMBL-Z86115; DDBJ-Z86115  
ENTRY DATE: Entered STN: 22 Jul 1998  
Last Updated on STN: 10 Sep 1998

AB From a genomic library of the industrially used strain *Lactobacillus delbrueckii* subsp. *lactis* DSM7290, a gene designated *arbZ* (869 bp; encoding a 33.5 kDa protein) was isolated by screening *E. coli* transformants for the ability to utilize the **beta-glucoside** arbutin. Out of 9000 transformants nine were able to ferment arbutin, whereas no utilization of the **beta-glucosides** salicin, esculin or cellobiose could be detected. Overexpression of *arbZ* using the T7-polymerase-T7-promoter-system resulted in the formation of insoluble, catalytically inactive protein aggregates (inclusion bodies). Accordingly, overexpression was not accompanied by an increase in *ArbZ* activity. Induction of *arbZ* controlled by the *lac* promoter under conditions that reduce protein aggregation resulted in a 12-fold increase in arbutin hydrolyzing activity of intact cells and a 13-fold increase in phospho-**beta-glycosidase** activity in cell-free extracts of the respective transformants. Nucleotide sequence analysis revealed a second gene upstream of *arbZ* that was designated *arbX* (830 bp). *ArbX* (32.6 kDa) shared similarity with several glycosyltransferases involved in the biosynthesis of lipopolysaccharides in Gram-negative bacteria. In *Lb. delbrueckii* subsp. *lactis* DSM7290 two transcripts, one covering *arbX* together with *arbZ* and one covering *arbZ* alone were detected by Northern blot analysis.

L10 ANSWER 3 OF 6 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN  
ACCESSION NUMBER: 1990:244076 BIOSIS  
DOCUMENT NUMBER: PREV199089131029; BA89:131029  
TITLE: INHIBITION OF CELL WALL-ASSOCIATED ENZYMES IN-VITRO AND IN-VIVO WITH SUGAR ANALOGS.  
AUTHOR(S): NAGAHASHI G [Reprint author]; TU S-I; FLEET G; NAMGOONG S K  
CORPORATE SOURCE: US DEP AGRIC, AGRIC RES SERVICE, EASTERN REGIONAL RES CENTER, 600 EAST MERMAID LANE, PHILADELPHIA, PA 19118, USA  
SOURCE: Plant Physiology (Rockville), (1990) Vol. 92, No. 2, pp. 413-418.  
CODEN: PLPHAY. ISSN: 0032-0889.  
DOCUMENT TYPE: Article  
FILE SEGMENT: BA  
LANGUAGE: ENGLISH  
ENTRY DATE: Entered STN: 19 May 1990  
Last Updated on STN: 19 May 1990

AB Sugar analogs were used to study the inhibition of cell wall-associated glycosidases in vitro and in vivo. For in vitro characterization, cell walls were highly purified from corn (*Zea mays* L.) root cortical cells and methods were developed to **assay** enzyme activity in situ. Inhibitor dependence curves, mode of inhibition and specificity were determined for three sugar analogs. At low concentrations of castanospermine (CAS), 2-acetamido-1,5-imino-1,2,5-trideoxy-D-glucitol, and swainsonine, these inhibitors showed competitive inhibition kinetics with  $\beta$ -glucosidase,  $\beta$ -GlcNAcase, and  $\alpha$ -mannosidase, respectively. Swainsonine specifically inhibited  $\alpha$ -mannosidase activity, and 2-acetamido-1,5-imino-1,2,5-trideoxy-D-glucitol specifically inhibited  $\beta$ -N-acetyl-hexosaminidase activity. However, CAS inhibited a broad spectrum of cell wall-associated enzymes. When the sugar analogs were applied to 2 day old corn seedlings, only CAS caused considerable changes in root growth and development. To ensure that the concentration of inhibitors used in vitro also inhibited enzyme activity in vivo, an in vivo method for measuring cell wall-associated activity was devised.

L10 ANSWER 4 OF 6 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1990:473466 HCAPLUS  
 DOCUMENT NUMBER: 113:73466  
 TITLE: The characteristics of  $\beta$ -glucosidase of chorionic villi and its fluorometric determination  
 AUTHOR(S): Wang, Jianzhi; He, Shanshu  
 CORPORATE SOURCE: Tonji Med. Univ., Wuhan, Peop. Rep. China  
 SOURCE: Shengwu Huaxue Zazhi (1990), 6(2), 102-6  
 CODEN: SHZAE4; ISSN: 1000-8543  
 DOCUMENT TYPE: Journal  
 LANGUAGE: Chinese  
 AB Acidic  $\beta$ -glucosidase (GBA) and neutral  $\beta$ -glucosidase (GBN) were isolated and purified from liver and kidney of rats by centrifugation and gel chromatog. **beta.-Glycosidase** of human chorionic villi showed similar electrophoretic behavior, pH relations, Km, and response to detergents (e.g., taurocholic acid and taurodeoxycholic acid) as GBA but not as GBN. Therefore,  $\beta$ -glucosidase of human chorionic villi was GBA. A fluorometric method using 4-methylumbelliferone . **beta.-glucoside** as substrate was developed for determination of GBA in chorionic villi of human. The average activity of GBA of chorionic villi from 76 cases of normal abortion was 263.7 nmol 4-methylumbelliferone/mg protein/h.

L10 ANSWER 5 OF 6 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1986:621623 HCAPLUS  
 DOCUMENT NUMBER: 105:221623  
 TITLE: Specificity of human glucosylceramide  $\beta$ -glucosidase towards synthetic glucosylsphingolipids inserted into liposomes. Kinetic studies in a detergent-free **assay** system  
 AUTHOR(S): Sarmientos, Francesco; Schwarzmann, Guenter; Sandhoff, Konrad  
 CORPORATE SOURCE: Inst. Org. Chem. Biochem., Univ. Bonn, Bonn, D-5300/1, Fed. Rep. Ger.  
 SOURCE: European Journal of Biochemistry (1986), 160(3), 527-35  
 CODEN: EJBCAI; ISSN: 0014-2956  
 DOCUMENT TYPE: Journal  
 LANGUAGE: English  
 AB The behavior of highly purified glucosylceramide  $\beta$ -glucosidase (EC 3.2.1.45) (I) from human placenta was investigated in the absence of detergents with structurally modified glucosylceramides inserted into unilamellar liposomes. The reaction between the water-soluble enzyme and the liposomal substrates was significantly dependent on the structure of the lipophilic aglycon moiety of glycolipids: glucosyl-N-acetylsphingosines (D-erythro and L-threo) were better than the corresponding glucosylceramides. The L-threo derivs. were poorer substrates with higher apparent Km values than the corresponding D-erythro derivs. For glucosyl-3-ketoceramide and glucosyldihydroceramide (D-erythro), higher Km values were found than for glucosylceramide. Sphingosine, glucosylsphingosine, and glucosyl-N-acetylsphingosine were the most effective inhibitors of the hydrolysis of glucosylceramide. D-erythro-Ceramide and D-galactosyl-N-acetyl-D-erythro-sphingosine inhibited the hydrolysis of amphiphilic glucosylceramide, but not that of water-soluble methylumbelliferyl-**beta.-glucoside**, suggesting a hydrophobic binding site of the enzyme for the aglycon moiety of its membrane-bound substrate. Dilution expts. suggested that at least a fraction of the enzyme assoc. with the liposomes and degrades the lipid substrate even in the absence of activator proteins. Acidic phospholipids incorporated into liposomes caused a powerful stimulation (30-40-fold) of the glucosylceramide  $\beta$ -glucosidase, whereas acidic sphingolipids (sulfatide, gangliosides GM1 and GD1a) incorporated into liposomes

stimulated this enzyme only moderately (3-10-fold).

L10 ANSWER 6 OF 6 HCAPLUS COPYRIGHT 2005 ACS on STN  
ACCESSION NUMBER: 1964:413184 HCAPLUS  
DOCUMENT NUMBER: 61:13184  
ORIGINAL REFERENCE NO.: 61:2225c-e  
TITLE: Fluorimetric determination of glycosidases in the  
locust (*Locusta migratoria*) and other insects  
AUTHOR(S): Robinson, D.  
CORPORATE SOURCE: Univ.London  
SOURCE: Comparative Biochemistry and Physiology (1964), 12(1),  
95-105  
CODEN: CBCPAI; ISSN: 0010-406X  
DOCUMENT TYPE: Journal  
LANGUAGE: Unavailable

AB Fluorimetric **assay** was found more suitable and sensitive than  
colorimetric or spectrophotometric methods for the determination of  
glycosidase  
activity in insects. The greatest activity was in the locust homogenates,  
the least in larvae and sap-sucking insects.  $\beta$ -Glucosidase and  
 $\beta$ -galactosidase were always present in ratios varying between 2:1 and  
1:2, with the exceptions of *Musca domestica* and *Blattella germanica* where  
the ratios were 1:8.8 and 5.6:1, resp. The  $\beta$ -glucuronidase activity  
was not inhibited by any of the glycosides and free sugars tested.  
Salicin and galactose inhibited  $\beta$ -galactosidase. Both  
 $\beta$ -glucosidase and  $\beta$ -galactosidase activities can function maximum  
in the presence of other substrate, suggesting no competition for the same  
active site; inhibition does take place when  $\alpha$ -arabinoside and  
**beta.-glucoside** are present at the same time. After  
heat treatment at 60° for 1 hr.,  $\beta$ -galactosidase lost less  
than half its activity while the half-life of  $\beta$ -glucosidase was 7  
min. Electrophoresis indicated that  $\beta$ -glucosidase in the locust is  
associated with at least 3 components only 1 of which had arabinosidase  
activity. It appears that  $\alpha$ -L-arabinosidase is distinct from  
 $\beta$ -galactosidase or  $\beta$ -glucosidase but that the arabinoside has an  
inhibitory effect on the latter. The function of the 3 components is  
unknown.

=> s hydrolysis

L11 786835 HYDROLYSIS

=> d his

(FILE 'HOME' ENTERED AT 10:44:36 ON 21 APR 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,  
LIFESCI' ENTERED AT 10:45:06 ON 21 APR 2005

L1 2266 S BETA(A) GLYCOSIDASE?  
L2 213 S ASSAY? AND L1  
L3 151781 S OLIGOSACCHARIDE? OR HETEROSACCHARIDE?  
L4 11 S L2 AND L3  
L5 7 DUP REM L4 (4 DUPLICATES REMOVED)  
L6 16 S ALKY(A) CHAIN?  
L7 0 S L2 AND L6  
L8 5827 S BETA(A) GLUCOSIDE  
L9 10 S L2 AND L8  
L10 6 DUP REM L9 (4 DUPLICATES REMOVED)  
L11 786835 S HYDROLYSIS

=> s l2 and l11

L12 23 L2 AND L11

=> dup rem l12  
PROCESSING COMPLETED FOR L12  
L13 14 DUP REM L12 (9 DUPLICATES REMOVED)

=> d 1-14 ibib ab

L13 ANSWER 1 OF 14 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on  
STN DUPLICATE 1  
ACCESSION NUMBER: 2005:308858 SCISEARCH  
THE GENUINE ARTICLE: 905LG  
TITLE: Determination of the hydrolytic activity of Achatina  
achatina beta-glucosidase toward some cyanogenic  
glycosides of some tropical plants  
AUTHOR: Okafor P N (Reprint)  
CORPORATE SOURCE: Fed Univ Agr, Dept Chem Sci, PMB 7267, Umudike, Umuahia  
Abia St, Nigeria (Reprint); Fed Univ Agr, Dept Chem Sci,  
Umudike, Umuahia Abia St, Nigeria  
COUNTRY OF AUTHOR: Nigeria  
SOURCE: PROCESS BIOCHEMISTRY, (APR 2005) Vol. 40, No. 5, pp.  
1579-1582.  
Publisher: ELSEVIER SCI LTD, THE BOULEVARD, LANGFORD LANE,  
KIDLINGTON, OXFORD OX5 1GB, OXON, ENGLAND.  
ISSN: 0032-9592.  
DOCUMENT TYPE: Article; Journal  
LANGUAGE: English  
REFERENCE COUNT: 15

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB The quantitative cyanide liberation resulting from **hydrolysis**  
of the cyanogenic glycosides of some edible tropical cyanophoric plants by  
a P-glucosidase enzyme prepared from African giant snail (Achatina  
achatina) was studied spectrophotometrically. The results were compared  
with that obtained with cassava linamarase that has relatively the same  
unit of activity. The quantitative yield of HCN equivalent from fresh  
cassava root pulp following incubation with A. achatina beta-glucosidase  
(4.7 units ml(-1)) was 1.65-14.46 mg CN(-)100 g(-1) fresh weight (for 10  
different cassava cultivars) and 1.67-14.17 mg CN- 100 g(-1) fresh weight  
when incubated with cassava linamarase (5.0 units ml(-1)) under the same  
**assay** conditions. There was no statistically significant  
differences (P > 0.01) between the hydrolytic activities of these two  
beta-glucosidases towards the cyanogenic glycosides of cassava, lima  
beans, wheat and standard linamarin. These results strongly indicate that  
A. achatina is another good source of P-glycosidase with high hydrolytic  
activity towards cyanogenic glycosides of some plants including cassava.  
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L13 ANSWER 2 OF 14 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN  
ACCESSION NUMBER: 2003-10795 BIOTECHDS  
TITLE: Novel isolated L-arabinose isomerase active enzyme derived  
from Thermoanaerobacter sp. capable of isomerizing  
D-galactose to D-tagatose, useful for producing D-tagatose;  
involving vector-mediated gene transfer and expression in  
host cell for use in aldose conversion and ketose and  
D-tagatose preparation  
AUTHOR: HANSEN O C; JORGENSEN F; STOUGAARD P; BERTELSEN H; BOTTCHER  
K; CHRISTENSEN H J S; ERIKNAUER K  
PATENT ASSIGNEE: BIOTEKNOLOGISK INST  
PATENT INFO: WO 2003008593 30 Jan 2003  
APPLICATION INFO: WO 2002-DK498 15 Jul 2002  
PRIORITY INFO: US 2001-905108 16 Jul 2001; US 2001-305155 16 Jul 2001  
DOCUMENT TYPE: Patent  
LANGUAGE: English  
OTHER SOURCE: WPI: 2003-239341 [23]  
AB DERWENT ABSTRACT:

NOVELTY - An isolated L-arabinose isomerase active enzyme (I) derived from a Thermoanaerobacter sp. or its active fragment, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following: (1) a L-arabinose isomerase (II) capable of isomerizing D-galactose to D-tagatose and having at least 70% sequence identity to a fully defined sequence of 465 amino acids (S1) as given in the specification, or its active fragment; (2) a nucleic acid (III) coding for L-arabinose isomerase or an L-arabinose isomerase active fragment, chosen from a wild type nucleic acid isolated from a Thermoanaerobacter sp., and a nucleic acid sequence that is capable of hybridizing with the sequence under stringent conditions; (3) a nucleic acid construct (IV) comprising (III); (4) a cell that is transformed with (III) or (IV); (5) preparation of (I); and (6) a composition comprising (I) in an immobilized form.

BIOTECHNOLOGY - Preparation: Producing (I) involves transforming a cell with (III) and operably linking to the nucleic acid, appropriate expression signals directing the expression of the isomerase and, optionally, sequences directing the secretion of the isomerase, propagating the transformed cell and harvesting the progeny cells containing the isomerase or if it is secreted into the medium, the excreted isomerase. The cell being transformed is a cell chosen from a bacterial cell, yeast cell and cell of a filamentous fungus. The method further involves purifying (I) from the progeny cells or the medium to obtain an L-arabinose isomerase preparation. The isomerase is purified to an extent where it is essentially without any other proteins. The method further comprises drying the preparation to a moisture content of at the most 10% by weight (claimed). Preferred Enzyme: (I) is derived from Thermoanaerobacter mathranii. (I) has at least one of the following characteristics: optimum activity at a temperature in the range of 60-80degreesC, optimum activity at a pH in the range of 7-9, and is capable of isomerizing at least one aldopentose and at least one aldohexose. Preferably, (I) is capable of isomerizing L-arabinose, D-galactose and D-fucose. (II) has at least 90% sequence identity to the sequence of (S1). Preferred Nucleic Acid: The wild type nucleic acid is isolated from Thermoanaerobacter mathranii, and codes for the amino acid sequence of (S1) or its active fragment. Preferred Construct: (IV) is chosen from a plasmid, chromosome, bacteriophage, transposon and cosmid.

USE - (I) is useful for converting an aldose into a ketose which involves contacting the aldose with (I), and keeping the reaction under conditions where at least 1% by weight of the aldose is converted. The aldose is chosen from L-arabinose, D-galactose and D-fucose, and the conversion reaction takes place at 60degreesC. At least 10% (preferably 25%) by weight of the substrate aldose is converted into its corresponding ketose, preferably D-tagatose. (I) is provided preferably as an immobilized isolated enzyme preparation. The method further comprises the step of hydrolyzing lactose to obtain D-galactose by use of a lactase-active enzyme, where D-galactose is converted to D-tagatose. The lactase is immobilized and the isomerase enzyme preparation is immobilized, and where the lactose **hydrolysis** and the aldose isomerization takes place in the same reactor. The lactase-active enzyme is preferably **beta-glycosidase**. (I) is useful for producing D-tagatose which involves hydrolyzing lactose by contacting lactose with a lactase-active enzyme to yield glucose and D-galactose, and converting at least a portion of the obtained D-galactose to D-tagatose by contacting the D-galactose with (I), where lactase-active enzyme and (I) are contained in the same reactor unit under essentially the same reaction conditions including a temperature in the range of about 60-100degreesC. The lactase-active enzyme and (I) are preferably immobilized. The reactions are carried out at a temperature in the range of about 60-80degreesC, including the range of about 65-80degreesC (claimed).

ADVANTAGE - (I) has a different substrate specificity as compared to other L-arabinose isomerases, and it is a versatile aldose isomerase

capable of isomerizing structurally related aldoses. (I) has maximum activity at around 65degreesC and about or more than 70% of the maximum activity is retained in the temperature range of about 60-75degreesC. Isomerization at high temperatures has the following advantages: (i) contamination risk from other microorganisms is minimized at such elevated temperatures; (ii) high temperatures allow the use of higher substrate concentrations due to increased substrate solubility; (iii) equilibrium between aldose and ketose is shifted towards ketose at higher temperatures; and (iv) substrate specificity towards D-galactose is higher relative to L-arabinose at higher temperatures.

EXAMPLE - Anaerobic, thermophilic microorganism *Thermoanaerobacter mathranii* DSMZ 11426 was cultivated at 65degreesC under anaerobic conditions. After growth, the culture was centrifuged and the pellet was stored at -80degreesC. Total chromosomal DNA was purified from frozen *T.mathranii* cells. Purified chromosomal DNA was partially cleaved with *Sau3A* restriction enzyme and DNA fragments of about 3-4 kb were purified. The pBluescript KS (+/-) plasmid was cleaved with *Bam*HI restriction enzyme, treated with alkaline phosphatase and purified from agarose gels. After ligation of purified DNA fragments and purified plasmid vector, the ligation mixture was introduced into supercompetent DH10B cells by electroporation. Transformed cells were plated onto Luria Bertani (LB) medium containing ampicillin (100 microg/ml). About 16,000 colonies were pooled from 20 plates and plasmid preparation from the pooled cells. The plasmid library prepared from pooled cells were introduced by electroporation into UP 1089 cells, an *Escherichia coli* strain carrying an *araA* mutation that prevents it from growing on an L-arabinose minimal medium. After transformation, the UP1089 cells were plated onto minimal medium containing L-arabinose as the only sugar, thus selected from complementation of the *araA* mutation. About 90 colonies were obtained by selection for growth on L-arabinose minimal medium plates. Plasmid inserts from two of these colonies were selected for DNA sequencing and they were found to contain identical DNA fragments comprising an L-arabinose isomerase gene (*araA*) followed by an L-ribulokinase gene (*araB*). The open reading frame of the *araA* gene encoded 465 amino acid residues, corresponding to a molecular weight of 52, 785 Da. L-arabinose isomerase activity was determined as described by Yamanaka, K. and Wood, W.A. (1996) *Methods in Enzymology* 9: 596-602. Enzyme sample, 10-50 microl, was mixed with 950 microl of **assay** reagent and incubated at 65degreesC for 60 min. The final concentrations were: L-arabinose, 5 mM; *MnCl*2, 5 mM; maleate buffer, pH 6.9, 25 mM. The obtained concentrations of the ketoses L-ribulose, D-tagatose or D-fuculose, respectively, were determined by the cysteine-carbozyl-sulfuric acid method Dische, Z. and Borenfreund, E. (1951) *J. Biol.*

#### Chemical

192: 583-587. D-galactose **assays** performed at increasing temperatures between 45degreesC and 85degreesC showed highest activity 65degreesC. One-ml **assay** mixtures containing 0.20 ml of *E.coli* cell extract with recombinant L-arabinose isomerase from *T.mathranii*, 0.30 g of D-galactose (30%, 1.67 M) or 0.60 g of D-galactose (60%, 3.33 M), 25 mM maleate buffer, pH 6.9 and 5 mM *MnCl*2 were incubated at 65degreesC. Control samples without enzyme were treated similarly. Periodically, samples were taken and the concentration of D-tagatose was determined. 26% bioconversion of D-galactose to D-tagatose was observed after 48 hours of incubation, with free enzyme (initial concentration of D-galactose 30%) while 19% bioconversion of D-galactose to D-tagatose was observed after 72 hours of incubation, with free enzyme (initial concentration of D-galactose 60%) (62 pages)

L13 ANSWER 3 OF 14 MEDLINE on STN DUPLICATE 2  
ACCESSION NUMBER: 2002155516 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 11835135  
TITLE: Screening of various glycosidases for the synthesis of octyl glucoside.

AUTHOR: Ducret Amelie; Trani Michael; Lortie Robert  
 CORPORATE SOURCE: Microbial and Enzyme Technology Group, Bioprocess Sector,  
 Biotechnology Research Institute, National Research  
 Council, Montreal, Quebec H4P 2R2 Canada.  
 SOURCE: Biotechnology and bioengineering, (2002 Mar 30) 77 (7)  
 752-7.  
 Journal code: 7502021. ISSN: 0006-3592.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200205  
 ENTRY DATE: Entered STN: 20020313  
 Last Updated on STN: 20020510  
 Entered Medline: 20020509

AB Thirteen glycosidases of microbial origin and almond **beta-glycosidase** were **assayed** in octanol/DMF (80:20, v/v), using a combination of **hydrolysis**, transglycosylation, and condensation reactions, in order to assess their potential for the production of alkyl glucosides. The two mesophile enzymes were highly impaired by the organic media. Three of the 11 thermophile enzymes gave interesting results in the **hydrolysis** and transglycosylation reactions, but they were highly inhibited by glucose. This made their use in a condensation reaction less interesting than the use of almond beta-glucosidase, which has a lower activity but shows less inhibition by the glucose.  
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 Biotechnol Bioeng 77: 752 757, 2002; DOI 10.1002/bit.10156

L13 ANSWER 4 OF 14 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 2001:825464 SCISEARCH  
 THE GENUINE ARTICLE: 480UA  
 TITLE: Production of a lactose-free galacto-oligosaccharide mixture by using selective enzymatic oxidation of lactose into lactobionic acid  
 AUTHOR: Splechtna B; Petzelbauer I; Baminger U; Haltrich D; Kulbe K D; Nidetzky B (Reprint)  
 CORPORATE SOURCE: Agr Univ Vienna, Inst Food Technol, Div Biochem Engn, Muthgasse 18, A-1190 Vienna, Austria (Reprint); Agr Univ Vienna, Inst Food Technol, Div Biochem Engn, A-1190 Vienna, Austria  
 COUNTRY OF AUTHOR: Austria  
 SOURCE: ENZYME AND MICROBIAL TECHNOLOGY, (4 OCT 2001) Vol. 29, No. 6-7, pp. 434-440.  
 Publisher: ELSEVIER SCIENCE INC, 655 AVENUE OF THE AMERICAS, NEW YORK, NY 10010 USA.  
 ISSN: 0141-0229.  
 DOCUMENT TYPE: Article; Journal  
 LANGUAGE: English  
 REFERENCE COUNT: 21

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB We report a novel and efficient way of producing lactose-derived galacto-oligosaccharides (GOS) that do not contain remaining lactose and monosaccharides. The initial sugar mixture was obtained by enzymatic transformation at 70 degreesC of a lactose solution of 270 g/liter using recombinant P-glycosidase from the Archaeon Sulfolobus solfataricus. At the optimum reaction time for kinetically controlled transgalactosylation, it contained 46% monosaccharides, 13% lactose and 41% GOS. Lactose was selectively oxidised into lactobionic acid by using fungal cellobiose dehydrogenase which displays a approximate to 100-fold preference for reaction with lactose compared to reaction with GOS. Oxidation of lactose



was coupled to reduction of 2,6-dichloro-indophenol which was added in catalytic concentrations. The oxidised redox mediator was regenerated continuously by fungal laccase-catalysed reduction of molecular oxygen into water. Ion exchange chromatographies were employed to remove lactobionic acid, other ions and monosaccharides. The final product contained 97% GOS, 1.2% lactose and 2.1% monosaccharides. The yield accounted for 25% of original lactose. An enzymatic **assay** for lactose has been developed. It is robust and allows sensitive quantification of the analyte in complex sugar mixtures containing large excesses of monosaccharides and GOS. (C) 2001 Elsevier Science Inc. All rights reserved.

L13 ANSWER 5 OF 14 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1999:639475 HCAPLUS  
DOCUMENT NUMBER: 132:1486  
TITLE: Probing the transferase activity of glycosidases by means of in situ NMR spectroscopy  
AUTHOR(S): Spangenberg, P.; Chiffolleau-Giraud, V.; Andre, C.; Dion, M.; Rabiller, C.  
CORPORATE SOURCE: Unite de Recherches en Biocatalyse (UPRES no. 2161), Faculte des Sciences et des Techniques, Nantes, F-44322, Fr.  
SOURCE: Tetrahedron: Asymmetry (1999), 10(15), 2905-2912  
CODEN: TASYE3; ISSN: 0957-4166  
PUBLISHER: Elsevier Science Ltd.  
DOCUMENT TYPE: Journal  
LANGUAGE: English

AB This paper describes the conditions in which in situ NMR spectroscopy is a suitable technique to use when following the course of enzymic transglycosylation reactions. Using this methodol., the reactions must be carried out in D2O. Our expts. indicate that the rate of the transglycosylation reaction is reduced in this solvent while the rate of the **hydrolysis** of the disaccharides produced is enhanced depending on the nature of the anomeric substituent. However, this undesirable effect is generally weak because the rates of the transglycosylation reactions are always faster than the rates of the **hydrolysis** whatever the solvent. The great advantage of NMR spectroscopy lies in its potential to detect, in a single experiment, all the

components of the reaction without any disturbance of the reaction medium.  
REFERENCE COUNT: 19 THERE ARE 19 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 6 OF 14 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1998:912037 SCISEARCH  
THE GENUINE ARTICLE: 141WE  
TITLE: Selenium-mediated differential response of beta-glucosidase and beta-galactosidase of germinating Trigonella foenum-graecum  
AUTHOR: Sreekala M; Lalitha K (Reprint)  
CORPORATE SOURCE: INDIAN INST TECHNOL, DEPT CHEM, MADRAS, CHENNAI, INDIA (Reprint); INDIAN INST TECHNOL, DEPT CHEM, MADRAS, CHENNAI, INDIA  
COUNTRY OF AUTHOR: INDIA  
SOURCE: BIOLOGICAL TRACE ELEMENT RESEARCH, (SUM 1998) Vol. 64, No. 1-3, pp. 247-258.  
Publisher: HUMANA PRESS INC, 999 RIVERVIEW DRIVE SUITE 208, TOTOWA, NJ 07512.  
ISSN: 0163-4984.  
DOCUMENT TYPE: Article; Journal  
FILE SEGMENT: LIFE  
LANGUAGE: English

REFERENCE COUNT: 35

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB beta-Glucosidase and beta-galactosidase activity profile tested in different seeds during 24 h germination revealed reasonably high levels of activity in *Vigna radiata*, *Cicer arietinum*, and *Trigonella foenum-graecum*. In all seeds tested, beta-galactosidase activity was, in general, higher than that of beta-glucosidase. *T. foenum-graecum* seedlings exhibited maximal total and specific activities for both the enzymes during 72 h germination. Se supplementation as Na<sub>2</sub>SeO<sub>3</sub> up to 0.75 ppm was found to be beneficial to growth and revealed selective enhancement of beta-galactosidase activity by 40% at 0.5 ppm Se. The activities of both the enzymes drastically decreased at 1.0 ppm level of Se supplementation. On the contrary, addition of Na<sub>2</sub>SeO<sub>3</sub> in vitro up to 1 ppm to the enzyme extracts did not influence these activities. Hydrolytic rates of beta-glucosidase in both control and Se-supplemented groups were enhanced by 20% with 0.05 M glycerol in the medium and 30% at 0.1 M glycerol. The rates were marginally higher in Se-supplemented seedlings than the controls, irrespective of added glycerol in the medium. In contrast, **hydrolysis** by beta-galactosidase showed a trend of decrease in Se-supplemented seedlings compared to the control, when glycerol was present in the medium. Addition of Se in vitro in the **assay** medium showed no difference in the hydrolytic rate by beta-galactosidase when compared to control, while the activity of beta-glucosidase declined by 50%. Se-grown seedlings showed an enhancement of transglucosidation rate by 40% in the presence of 0.1 M glycerol. The study reveals a differential response to Se among the beta-galactosidase and beta-glucosidase of *T. foenum-graecum* with increase in the levels of beta-galactosidase activity.

L13 ANSWER 7 OF 14 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1998:214759 SCISEARCH

THE GENUINE ARTICLE: ZB029

TITLE: **beta-glycosidase** (amygdalase and linamarase) from *Endomyces fibuliger* (LU677): formation and crude enzyme properties

AUTHOR: Brimer L (Reprint); Nout M J R; Tuncel G

CORPORATE SOURCE: ROYAL VET & AGR UNIV, DEPT PHARMACOL & PATHOBIOL, SECT PHARMACOL & TOXICOL, 13 BULOWSVEJ, DK-1870 FREDERIKSBURG C, DENMARK (Reprint); UNIV AGR, DEPT FOOD TECHNOL & NUTR SCI, NL-6703 HD WAGENINGEN, NETHERLANDS; EGE UNIV, FAC ENGN, TR-35100 IZMIR, TURKEY

COUNTRY OF AUTHOR: DENMARK; NETHERLANDS; TURKEY

SOURCE: APPLIED MICROBIOLOGY AND BIOTECHNOLOGY, (FEB 1998) Vol. 49, No. 2, pp. 182-188.

Publisher: SPRINGER VERLAG, 175 FIFTH AVE, NEW YORK, NY 10010.

ISSN: 0175-7598.

DOCUMENT TYPE: Article; Journal

FILE SEGMENT: LIFE; AGRI

LANGUAGE: English

REFERENCE COUNT: 39

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB In our previous studies, the yeast *Endomyces fibuliger* LU677 was found to degrade amygdalin in bitter apricot seeds. The present investigation shows that *E. fibuliger* LU677 produces extracellular **beta-glycosidase** activity when grown in malt extract broth (MEB). Growth was very good at 25 degrees C and 30 degrees C and slightly less at 35 degrees C. When grown in MEB of pH 5 and pH 6 with addition of 0, 10 or 100 ppm amygdalin, *E. fibuliger* produced only slightly more biomass at pH 5, and was only slightly inhibited in the presence of amygdalin. Approximately, 60% of the added amygdalin was degraded (fastest at 35 degrees C) during an incubation period of 5 days. Supernatants of cultures

grown at 25 degrees C and pH 6 for 5 days were tested for the effects of pH and temperature on activity (using amygdalin, linamarin and prunasin as substrates). Prunase activity had two pH optima (pH 4 and pH 6), amygdalase and linamarase only one each at pH 6 and pH 4-5 respectively. The linamarase activity evolved earlier than amygdalase (2 days and 4 days respectively). The data thus indicate the presence of at least two different glycosidases having different pH optima and kinetics of excretion. In the presence of amygdalin, lower glycosidase activities were generally produced. However, the amygdalin was degraded from the start of the growth, strongly indicating an uptake of amygdalin by the cells. The temperature optimum for all activities was at 40 degrees C. Activities of amygdalase (**assayed** at pH 4) and linamarase (at pH 6) evolving during the growth of *E. fibuliger* were generally higher in cultures grown at 25 degrees C and 30 degrees C. TLC analysis of amygdalin degradation products show a two-stage sequential mechanism as follows: (1) amygdalin to prunasin and (2) prunasin to cyanohydrin.

L13 ANSWER 8 OF 14 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1996:703733 HCAPLUS

DOCUMENT NUMBER: 126:18002

TITLE: Molecular basis of alcoholic aroma formation in tea leaves

AUTHOR(S): Sakata, Kanzo; Ogawa, Kenji; Ijima, Yasuyuki; Watanabe, Naoharu; Usui, Taiichi; Guo, Wenfei; Moon, Jae-Hak; Dong, Shangsheng; Tong, Qiqing; Luo, Shaojun

CORPORATE SOURCE: Faculty Agriculture, Shizuoka University, Japan

SOURCE: Tennen Yuki Kagobutsu Toronkai Koen Yoshishu (1996), 38th, 511-516

CODEN: TYKYDS

PUBLISHER: Nippon Kagakkai

DOCUMENT TYPE: Journal

LANGUAGE: English

AB In the course of studies on aroma formation mechanism in oolong tea (*Camellia sinensis sinensis*), the mol. basis was shown in that most of alc. tea aroma is generated by endogenous enzymic **hydrolysis** of glycosidic aroma precursors during tea manufacturing. The main alc. aroma precursors of geraniol, linalool, 2-phenylethanol, benzyl alc., linalool oxides I and II (trans- and cis-linalool 3,6-oxides) and Me salicylate as  $\beta$ -primeverosides, guided by an enzymic **hydrolysis** (with the acetone powder prepared from cv. Yabukita) followed by GC and GC-MS analyses were isolated and identified. Aroma precursors of linalool oxides III and IV (trans- and cis-linalool 3,7-oxides) and (Z)-3-hexenol were exceptionally isolated as 6-O- $\beta$ -D-apiofuranosyl- $\beta$ -D-glucopyranosides and a  $\beta$ -D-glucopyranoside, resp. from oolong tea leaves. In cv. Yabukita for green tea, benzyl alc. was stored as ca. 1:1 mixture of  $\beta$ -primeveroside and  $\beta$ -D-glucopyranoside. As a preliminary experiment, a new **glycosidase** (**.beta** **-primeverosidase**) was purified from fresh leaves of cv. Yabukita by ammonium sulfate precipitation followed by chromatogs. with CM Toyopearl and Mono

S-MR. This glycosidase was one of the main glycosidases involved in the alc. aroma formation in tea leaves. This is the 2nd example of  $\beta$ -primeverosidase. Purification of the  $\beta$ -primeverosidase from fresh oolong tea leaves was also carried out in the same manner. The mol. weight was shown lobe 61 and 60.3 kDa by SDS-PAGE and TGFMS, resp. Its pI and Optimum temperature and pM are 9.5, 45°C and 4, resp. The enzyme is stable below 45°C, and between pH 3 to 5. These enzymic characteristics are quite similar to those of the  $\beta$ -primeverosidase from cv. Yabukita. The  $\beta$ -primeverosidase hydrolyzed a  $\beta$ -apiofuranoside into each constituent dissacharide and aglycon without further **hydrolysis** as effectively as in the case of a  $\beta$ -primeverosidase. Amts. of alc. aroma precursor and glycosidase activity in each part of the tea shoot (cvs. Yabukita and Irumi) were

indirectly measured by means of a crude enzyme **assay**. The aroma precursors were abundant in young leaves and decreased as the leaf aged. Glycosidase activity also decreased as leaves aged, but was high in stems.

L13 ANSWER 9 OF 14 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1993:622449 HCAPLUS  
DOCUMENT NUMBER: 119:222449  
TITLE: Variable expression of leukocyte cytosolic broad-specificity  $\beta$ -glucosidase activity  
AUTHOR(S): Forsyth, G. W.; Romero, K. M.; Alverson, J.; VanderJagt, D. J.; Glew, R. H.  
CORPORATE SOURCE: Univ. Saskatchewan, Saskatoon, SK, S7N 0W0, Can.  
SOURCE: Clinica Chimica Acta (1993), 216(1-2), 11-21  
CODEN: CCATAR; ISSN: 0009-8981  
DOCUMENT TYPE: Journal  
LANGUAGE: English

AB The cytosolic  $\beta$ -glucosidase activity that is found in a variety of mammalian tissues has no clearly defined function. In vitro **assay** conditions under which the broad-specificity  $\beta$ -glucosidase hydrolyzes glucocerebroside at a significant rate have not been described. Nonetheless, it has been suggested that this enzyme might play an accessory role with lysosomal glucocerebrosidase in catalyzing the **hydrolysis** of glucosylceramide. However, this hypothesis would require that activity of both enzymes be low in severe cases of Gaucher disease in which there are pathol. accumulations of glucosylceramide in one or more of the affected organs, i.e. spleen, liver and bone marrow. Information is lacking regarding the normal range of cytosolic **beta.-glycosidase** activity in humans. Nitrophenyl- $\beta$ -D-mannoside was a potent inhibitor ( $K_i = 0.068$  mM) of cytosolic  $\beta$ -glucosidase. In parallel studies, the activity of glucocerebrosidase was minimally affected by nitrophenyl- $\beta$ -D-mannoside at concns. as high as 2.5 mM. This information was used to design an **assay** system that would allowed an estimate of glucocerebrosidase and cytosolic  $\beta$ -glucosidase activities in exts. of human leukocyte. The average cytosolic  $\beta$ -glucosidase activity with 4-heptyl-umbelliferyl- $\beta$ -D-glucose as a substrate was 8.8 nmol/h/mg protein in leukocytes from 356 subjects. The average leukocyte glucocerebrosidase specific activity was 16 nmol/h/mg protein. No correlation was observed between cytosolic  $\beta$ -glucosidase and glucocerebrosidase activity for control and Gaucher heterozygote populations. The wide range of leukocyte cytosolic  $\beta$ -glucosidase activity in individuals tested in this study indicates that a substantial proportion of the population may lack sufficient cytosolic  $\beta$ -glucosidase activity to assist a defective lysosomal glucocerebrosidase in hydrolyzing glucosylceramide.

L13 ANSWER 10 OF 14 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1990:194262 HCAPLUS  
DOCUMENT NUMBER: 112:194262  
TITLE:  $\beta$ -D-Glycosidase activities of Humicola grisea: biochemical and kinetic characterization of a multifunctional enzyme  
AUTHOR(S): Peralta, Rosane Marina; Terenzi, Hector Francisco; Jorge, Joao Atilio  
CORPORATE SOURCE: Fac. Filos. Cienc. Let., Univ. Sao Paulo, Ribeirao Preto, 14049, Brazil  
SOURCE: Biochimica et Biophysica Acta (1990), 1033(3), 243-9  
CODEN: BBACAQ; ISSN: 0006-3002  
DOCUMENT TYPE: Journal  
LANGUAGE: English

AB A  $\beta$ -D-glycosidase was purified from mycelium of *H. grisea* var. *thermoidea* grown on avicel as the main C source. The purified enzyme was a glycoprotein and migrated as a single polypeptide band on PAGE under

native or denaturing conditions. The apparent mol. weight of the enzyme was .apprx.55 kDa by gel filtration and SDS-PAGE. The enzyme was active against o-nitrophenyl  $\beta$ -D-galactoside; p-nitrophenyl  $\beta$ -D-glucoside, p-nitrophenyl  $\beta$ -D-fucoside, lactose, and cellobiose, PNP fucoside (synthetic substrate) and cellobiose (natural substrate) being the best utilized. A comparison of the properties of  $\beta$ -D-galactosidase,  $\beta$ -D-glucosidase, and  $\beta$ -D-fucosidase showed that 3 activities exhibited similar pH and temperature optima and the same thermostability. The **hydrolysis** rate of substrate mixts. suggests that the enzyme possesses a common catalytic site for all the substrates **assayed**.

L13 ANSWER 11 OF 14 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1986:621623 HCAPLUS

DOCUMENT NUMBER: 105:221623

TITLE: Specificity of human glucosylceramide  $\beta$ -glucosidase towards synthetic glucosylsphingolipids inserted into liposomes. Kinetic studies in a detergent-free **assay** system

AUTHOR(S): Sarmientos, Francesco; Schwarzmann, Guenter; Sandhoff, Konrad

CORPORATE SOURCE: Inst. Org. Chem. Biochem., Univ. Bonn, Bonn, D-5300/1, Fed. Rep. Ger.

SOURCE: European Journal of Biochemistry (1986), 160(3), 527-35

CODEN: EJBCAI; ISSN: 0014-2956

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The behavior of highly purified glucosylceramide  $\beta$ -glucosidase (EC 3.2.1.45) (I) from human placenta was investigated in the absence of detergents with structurally modified glucosylceramides inserted into unilamellar liposomes. The reaction between the water-soluble enzyme and the

liposomal substrates was significantly dependent on the structure of the lipophilic aglycon moiety of glycolipids: glucosyl-N-acetylsphingosines (D-erythro and L-threo) were better than the corresponding glucosylceramides. The L-threo derivs. were poorer substrates with higher apparent Km values than the corresponding D-erythro derivs. For glucosyl-3-ketoceramide and glucosyldihydroceramide (D-erythro), higher Km values were found than for glucosylceramide. Sphingosine, glucosylsphingosine, and glucosyl-N-acetylsphingosine were the most effective inhibitors of the **hydrolysis** of glucosylceramide. D-erythro-Ceramide and D-galactosyl-N-acetyl-D-erythro-sphingosine inhibited the **hydrolysis** of amphiphilic glucosylceramide, but not that of water-soluble methylumbelliferyl- $\beta$ -glucoside, suggesting a hydrophobic binding site of the enzyme for the aglycon moiety of its membrane-bound substrate. Dilution expts. suggested that at least a fraction

of the enzyme assoc. with the liposomes and degrades the lipid substrate even in the absence of activator proteins. Acidic phospholipids incorporated into liposomes caused a powerful stimulation (30-40-fold) of the glucosylceramide  $\beta$ -glucosidase, whereas acidic sphingolipids (sulfatide, gangliosides GM1 and GD1a) incorporated into liposomes stimulated this enzyme only moderately (3-10-fold).

L13 ANSWER 12 OF 14 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1986:182111 HCAPLUS

DOCUMENT NUMBER: 104:182111

TITLE: A fluorimetric method for measuring the activity of soil enzymes

AUTHOR(S): Darrah, P. R.; Harris, P. J.

CORPORATE SOURCE: Dep. Soil Sci., Univ. Reading, Reading, RG1 5AQ, UK

SOURCE: Plant and Soil (1986), 92(1), 81-8  
CODEN: PLSOA2; ISSN: 0032-079X  
DOCUMENT TYPE: Journal  
LANGUAGE: English

AB A fluorometric method is described for the measurement of the activity of a range of soil enzymes. The method is based on the measurement of 4-methylumbelliferone (MUB), a fluorescent product liberated on **hydrolysis** of the enzyme substrate. The main advantage of the method over colorimetric techniques is that separation of MUB from the soil is unnecessary and the method is therefore suitable for routine, automated analyses. The method was used to measure the activity of  $\beta$ -cellobiase,  $\beta$ -acetylgalactosaminidase,  $\beta$ -glucosidase, and  $\beta$ -xylosidase over a wide range of substrate concentration and in a range of soils. Kinetic parameters are reported for these enzymes. The method was also suitable for the **assay** of arylsulfatase and acid and alkaline phosphatases in soil. The technique should be applicable to a wide range of soil hydrolases, by using the same **assay** methods.

L13 ANSWER 13 OF 14 MEDLINE on STN DUPLICATE 3  
ACCESSION NUMBER: 79172457 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 108231  
TITLE: Hydrolase activities in normoblasts of beta-thalassemic patients.  
AUTHOR: Yatziv S; Abeliuk P; Rachmilewitz E A; Cividalli G; Kahane I  
SOURCE: Israel journal of medical sciences, (1978 Nov) 14 (11) 1124-6.  
Journal code: 0013105. ISSN: 0021-2180.  
PUB. COUNTRY: Israel  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 197907  
ENTRY DATE: Entered STN: 19900315  
Last Updated on STN: 19900315  
Entered Medline: 19790716

AB A physiological role for glycosidases in cell membranes has been suggested. Therefore the activities of four **glycosidases**--**beta**-galactosidase, beta-glucoronidase, N-acetyl-beta-glucosaminidase and acid phosphatase--were examined in normoblasts and membranes of red blood cells (RBC). The enzymatic **assays** were based on the **hydrolysis** of fluorimetric 4-methylumbelliferone from the enzyme substrate. In order to avoid contamination by lysosomal activities derived from RBC, the mature RBC and normoblasts obtained from normal controls and thalassemic patients were separated from other blood elements by cellulose chromatography. The cells were disrupted and lysed by freezing and thawing hypotonic solution. Higher enzymatic activities were found in preparations from thalassemic patients than from normal subjects. With a sucrose density gradient, further separation of normoblasts from RBC membranes was obtained, indicating that the normoblast fraction contributed most of the high specific activity found in the thalassemic preparation. It was concluded that relatively high glycosidase activities are present in normoblasts of thalassemic patients. Lower but significant activities were detected in RBC membranes of normal control subjects and thalassemic patients.

L13 ANSWER 14 OF 14 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN DUPLICATE 4  
ACCESSION NUMBER: 1978:235939 BIOSIS  
DOCUMENT NUMBER: PREV197866048436; BA66:48436  
TITLE: SCREENING FOR FLAVONOL 3 GLYCOSIDE SPECIFIC **BETA GLYCOSIDASES** IN PLANTS USING A SPECTROPHOTOMETRIC

ENZYMATIC **ASSAY**.  
 AUTHOR(S): SURHOLT E [Reprint author]; HOESEL W  
 CORPORATE SOURCE: UNIV HINDENBURGPL 55, D-4400 MUENSTER, W GER  
 SOURCE: Phytochemistry (Oxford), (1978) Vol. 17, No. 5, pp.  
 873-878.  
 CODEN: PYTCAS. ISSN: 0031-9422.  
 DOCUMENT TYPE: Article  
 FILE SEGMENT: BA  
 LANGUAGE: ENGLISH

AB A simple and rapid spectrophotometric **assay** for following the  
**hydrolysis** of flavonol 3-glycosides was developed. The  
**assay** profits from the fact that peroxidase converts flavonol  
 aglycones to their corresponding 2,3-dihydroxyflavanones, producing a  
 large shift in UV absorption, whereas flavonol 3-glycosides are not  
 attacked. The amount of liberated aglycone can therefore be calculated  
 from the decrease of flavonol absorption at 350-380 nm. A horseradish  
 peroxidase-H2O2 test system was used to investigate the **hydrolysis**  
 of most flavonol 3-glycosides, whereas quercetin 3-glycosides was tested  
 using a peroxidase preparation from Mentha sp. which uses O2 as cofactor  
 rather than H2O2. Flavonol 3-glycoside synthesis, e.g., with UDP-sugars  
 as cofactors, was also tested by this particular system. Various plants  
 and plant cell cultures were screened for kaempferol and quercetin  
 3-glycoside specific **.beta.-glycosidases**. In no case  
 could any specific activity be detected.

=> d his

(FILE 'HOME' ENTERED AT 10:44:36 ON 21 APR 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,  
 LIFESCI' ENTERED AT 10:45:06 ON 21 APR 2005

L1 2266 S BETA(A) GLYCOSIDASE?  
 L2 213 S ASSAY? AND L1  
 L3 151781 S OLIGOSACCHARIDE? OR HETEROSACCHARIDE?  
 L4 11 S L2 AND L3  
 L5 7 DUP REM L4 (4 DUPLICATES REMOVED)  
 L6 16 S ALKY(A)CHAIN?  
 L7 0 S L2 AND L6  
 L8 5827 S BETA(A)GLUCOSIDE  
 L9 10 S L2 AND L8  
 L10 6 DUP REM L9 (4 DUPLICATES REMOVED)  
 L11 786835 S HYDROLYSIS  
 L12 23 S L2 AND L11  
 L13 14 DUP REM L12 (9 DUPLICATES REMOVED)

=> e matsui I/au

E1 1 MATSUI HOROYUKI/AU  
 E2 1 MATSUI HROSHI/AU  
 E3 651 --> MATSUI I/AU  
 E4 1 MATSUI I G/AU  
 E5 1 MATSUI I KOJI/AU  
 E6 1 MATSUI I M/AU  
 E7 1 MATSUI I MASANAO/AU  
 E8 1 MATSUI I S/AU  
 E9 3 MATSUI I S L/AU  
 E10 1 MATSUI ICHINOSUKE/AU  
 E11 93 MATSUI ICHIRO/AU  
 E12 1 MATSUI ICHIZO/AU

=> s e3

L14 651 "MATSUI I"/AU

=> e ishikawa k/au

E1	1	ISHIKAWA JYUNKO/AU
E2	1	ISHIKAWA JYUZABURO/AU
E3	8696 -->	ISHIKAWA K/AU
E4	5	ISHIKAWA K */AU
E5	18	ISHIKAWA K B/AU
E6	1	ISHIKAWA K H/AU
E7	262	ISHIKAWA K I/AU
E8	1	ISHIKAWA K J/AU
E9	7	ISHIKAWA K L/AU
E10	1	ISHIKAWA K N/AU
E11	4	ISHIKAWA K S/AU
E12	4	ISHIKAWA K Y/AU

=> s e3

L15 8696 "ISHIKAWA K"/AU

=> e ishida h/au

E1	6	ISHIDA GORO/AU
E2	1	ISHIDA GOROU/AU
E3	6232 -->	ISHIDA H/AU
E4	1	ISHIDA H */AU
E5	1	ISHIDA H HIDEKI/AU
E6	4	ISHIDA H I/AU
E7	18	ISHIDA H K/AU
E8	2	ISHIDA H S/AU
E9	1	ISHIDA H Y/AU
E10	1	ISHIDA HAITOSHI/AU
E11	192	ISHIDA HAJIME/AU
E12	1	ISHIDA HAKASE/AU

=> s e3

L16 6232 "ISHIDA H"/AU

=> e kosugi y/au

E1	2	KOSUGI W/AU
E2	3	KOSUGI WATARU/AU
E3	467 -->	KOSUGI Y/AU
E4	1	KOSUGI Y */AU
E5	7	KOSUGI YAE/AU
E6	10	KOSUGI YASUHIKO/AU
E7	1	KOSUGI YASUHIRO/AU
E8	5	KOSUGI YASUHITO/AU
E9	1	KOSUGI YASUMASA/AU
E10	1	KOSUGI YASUNORI/AU
E11	1	KOSUGI YASUO/AU
E12	2	KOSUGI YASUSHI/AU

=> s e3

L17 467 "KOSUGI Y"/AU

=> d his

(FILE 'HOME' ENTERED AT 10:44:36 ON 21 APR 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 10:45:06 ON 21 APR 2005

L1	2266 S BETA(A) GLYCOSIDASE?
L2	213 S ASSAY? AND L1
L3	151781 S OLIGOSACCHARIDE? OR HETEROSACCHARIDE?
L4	11 S L2 AND L3
L5	7 DUP REM L4 (4 DUPLICATES REMOVED)
L6	16 S ALKY(A) CHAIN?



L7 0 S L2 AND L6  
 L8 5827 S BETA(A)GLUCOSIDE  
 L9 10 S L2 AND L8  
 L10 6 DUP REM L9 (4 DUPLICATES REMOVED)  
 L11 786835 S HYDROLYSIS  
 L12 23 S L2 AND L11  
 L13 14 DUP REM L12 (9 DUPLICATES REMOVED)  
 E MATSUI I/AU  
 L14 651 S E3  
 E ISHIKAWA K/AU  
 L15 8696 S E3  
 E ISHIDA H/AU  
 L16 6232 S E3  
 E KOSUGI Y/AU  
 L17 467 S E3

=> s l14 or l15 or l16 or l17  
 L18 15871 L14 OR L15 OR L16 OR L17

=> s l1 and l18  
 L19 6 L1 AND L18

=> dup rem l19  
 PROCESSING COMPLETED FOR L19  
 L20 2 DUP REM L19 (4 DUPLICATES REMOVED)

=> d 1-2 ibib ab

L20 ANSWER 1 OF 2 EMBASE COPYRIGHT 2005 ELSEVIER INC. ALL RIGHTS RESERVED.  
 on STN DUPLICATE 1

ACCESSION NUMBER: 2004397052 EMBASE  
 TITLE: X-ray structure of a membrane-bound **.beta.-glycosidase** from the hyperthermophilic archaeon *Pyrococcus horikoshii*.  
 AUTHOR: Akiba T.; Nishio M.; **Matsui I.**; Harata K.  
 CORPORATE SOURCE: K. Harata, Biol. Information Research Center, Natl. Inst. Adv. Indust. Sci./Tech., Central 6, 1-1-1 Higashi, Tsukuba, Ibaraki 305-8566, Japan. k-harata@aist.go.jp  
 SOURCE: Proteins: Structure, Function and Genetics, (1 Nov 2004)  
 Vol. 57, No. 2, pp. 422-431.  
 Refs: 42  
 ISSN: 0887-3585 CODEN: PSFGEY  
 COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article  
 FILE SEGMENT: 004 Microbiology  
 029 Clinical Biochemistry  
 LANGUAGE: English  
 SUMMARY LANGUAGE: English  
 ENTRY DATE: Entered STN: 20040930  
 Last Updated on STN: 20040930

AB The **.beta.-glycosidase** of the hyperthermophilic Archaeon *Pyrococcus horikoshii* is a membrane-bound enzyme with the preferred substrate of alkyl- $\beta$ -glycosides. In this study, the unusual structural features that confer the extreme thermostability and substrate preferences of this enzyme were investigated by X-ray crystallography and docking simulation. The enzyme was crystallized in the presence of a neutral surfactant, and the crystal structure was solved by the molecular replacement method and refined at 2.5 Å. The main-chain fold of the enzyme belongs to the ( $\beta\alpha$ ) (8) barrel structure common to the Family 1 glycosyl hydrolases. The active site is located at the center of the C-termini of the barrel  $\beta$ -strands. The deep pocket of the active site accepts one sugar unit, and a hydrophobic channel extending radially from there binds the nonsugar moiety of the substrate.

The docking simulation for oligosaccharides and alkylglucosides indicated that alkylglucosides with a long aliphatic chain are easily accommodated in the hydrophobic channel. This sparingly soluble enzyme has a cluster of hydrophobic residues on its surface, situated at the distal end of the active site channel and surrounded by a large patch of positively charged residues. We propose that this hydrophobic region can be inserted into the membrane while the surrounding positively charged residues make favorable contacts with phosphate groups on the inner surface of the membrane. The enzyme could thus adhere to the membrane in the proximity of its glycolipid substrate. .COPYRGT. 2004 Wiley-Liss, Inc.

L20 ANSWER 2 OF 2 MEDLINE on STN DUPLICATE 2  
 ACCESSION NUMBER: 2000141228 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 10675537  
 TITLE: Novel substrate specificity of a membrane-bound  
**beta-glycosidase** from the  
 hyperthermophilic archaeon *Pyrococcus horikoshii*.  
 AUTHOR: **Matsui I**; Sakai Y; Matsui E; Kikuchi H;  
 Kawarabayashi Y; Honda K  
 CORPORATE SOURCE: National Institute of Bioscience and Human-Technology,  
 Tsukuba, Ibaraki, Japan.. ikmatsui@nibh.go.jp  
 SOURCE: FEBS letters, (2000 Feb 11) 467 (2-3) 195-200.  
 Journal code: 0155157. ISSN: 0014-5793.  
 PUB. COUNTRY: Netherlands  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200003  
 ENTRY DATE: Entered STN: 20000413  
 Last Updated on STN: 20000413  
 Entered Medline: 20000331

AB A **beta-glycosidase** gene homolog of *Pyrococcus horikoshii* (BGPh) was successfully expressed in *Escherichia coli*. The enzyme was localized in a membrane fraction and solubilized with 2.5% Triton X-100 at 85 degrees C for 15 min. The optimum pH was 6.0 and the optimum temperature was over 100 degrees C, respectively. BGPh stability was dependent on the presence of Triton X-100, the enzyme's half-life at 90 degrees C (pH 6.0) was 15 h. BGPh has a novel substrate specificity with k(cat)/K(m) values high enough for hydrolysis of beta-D-Glcp derivatives with long alkyl chain at the reducing end and low enough for the hydrolysis of beta-linked glucose dimer more hydrophilic than aryl- or alkyl-beta-D-Glcp.

=> d his

(FILE 'HOME' ENTERED AT 10:44:36 ON 21 APR 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 10:45:06 ON 21 APR 2005

L1 2266 S BETA(A) GLYCOSIDASE?  
 L2 213 S ASSAY? AND L1  
 L3 151781 S OLIGOSACCHARIDE? OR HETEROSACCHARIDE?  
 L4 11 S L2 AND L3  
 L5 7 DUP REM L4 (4 DUPLICATES REMOVED)  
 L6 16 S ALKY(A) CHAIN?  
 L7 0 S L2 AND L6  
 L8 5827 S BETA(A) GLUCOSIDE  
 L9 10 S L2 AND L8  
 L10 6 DUP REM L9 (4 DUPLICATES REMOVED)  
 L11 786835 S HYDROLYSIS  
 L12 23 S L2 AND L11  
 L13 14 DUP REM L12 (9 DUPLICATES REMOVED)

L14           E MATSUI I/AU  
651 S E3  
L15           E ISHIKAWA K/AU  
8696 S E3  
L16           E ISHIDA H/AU  
6232 S E3  
L17           E KOSUGI Y/AU  
467 S E3  
L18           15871 S L14 OR L15 OR L16 OR L17  
L19           6 S L1 AND L18  
L20           2 DUP REM L19 (4 DUPLICATES REMOVED)

	Issue Date	Pages	Document ID	Title
1	19730327	4	US 3723506 A	OXIDATION OF SUGARS

	Issue Date	Pages	Document ID	Title
1	20050203	21	US 20050026249 A1	Enzymatically active recombinant glucocerebrosidase
2	20050127	21	US 20050019861 A1	Enzymatically active recombinant glucocerebrosidase
3	20041118	103	US 20040229830 A1	Delivery of therapeutic compositions using ultrasound
4	20040325	90	US 20040059313 A1	Ultrasound assembly for use with light activated drugs
5	20040212	17	US 20040028671 A1	Ginsenoside glycosidases hydrolyzing ginsenoside sugar moieties and uses thereof
6	20030821	90	US 20030157024 A1	ULTRASOUND ASSEMBLY FOR USE WITH LIGHT ACTIVATED DRUGS
7	20030515	103	US 20030092667 A1	Delivery of therapeutic compositions using ultrasound
8	20030102	14	US 20030003120 A1	Phloridzin-rich phenolic fraction and use thereof as a cosmetic, dietary or nutraceutical agent
9	20021114	21	US 20020168750 A1	Enzymatically active recombinant glucocerebrosidase
10	20020314	20	US 20020031796 A1	Method and composition for detecting bacterial contamination in food products
11	20041102	12	US 6811804 B2	Juice and soy protein beverage and uses thereof
12	20030520	38	US 6566585 B1	Genetically modified plant cells and plants with an increased activity of an amylosucrase protein and a branching enzyme
13	20030304	89	US 6527759 B1	Ultrasound assembly for use with light activated drugs

	Issue Date	Pages	Document ID	Title
14	20021029	9	US 6472167 B1	Method and composition for detecting bacterial contamination in food products
15	20020917	19	US 6451600 B1	Enzymatically active recombinant glucocerebrosidase
16	20020813	25	US 6433249 B1	Use of .beta.-glucosidase to enhance disease resistance and resistance to insects in crop plants
17	20020604	19	US 6399579 B1	Compositions comprising icariside I and anhydroicaritin and methods for making the same
18	20020514	11	US 6387650 B1	Method and composition for detecting bacterial contamination in food products
19	20020416	7	US 6372894 B1	Cycloalkylated B-glucoside
20	20020115	7	US 6339146 B1	Cycloalkylated beta-glucoside
21	20011211	24	US 6329208 B1	Methods for determining gluconeogenesis, anapleurosis and pyruvate recycling
22	20010123	89	US 6176842 B1	Ultrasound assembly for use with light activated drugs
23	19980505	14	US 5747320 A	Glucose and cellobiose tolerant .beta.-glucosidase from Candida peltata
24	19970311	12	US 5610029 A	Medium for detecting target microbes in a sample
25	19970225	6	US 5606035 A	Sesaminol glucosides
26	19961119	11	US 5576216 A	Universal standard reagents, method of preparing same and use thereof

	Issue Date	Pages	Document ID	Title
27	19960409	39	US 5506211 A	Genistein for use in inhibiting osteoclasts
28	19930817	21	US 5236838 A	Enzymatically active recombinant glucocerebrosidase
29	19830301	8	US 4375399 A	Molecule selective sensor for industrial use and procedure for its preparation
30	19821019	10	US 4354913 A	Molecule selective enzyme electrode
31	19730327	4	US 3723506 A	OXIDATION OF SUGARS
32	19720425	3	US 3658588 A	PREPARATION OF STARCH FROM CELLULOSE TREATED WITH PHOSPHORIC ACID

	L #	Hits	Search Text
1	L1	0	beat adj glycosidase\$2
2	L2	155	beta adj glycosidase\$2
3	L3	471	beta adj glucoside\$2
4	L4	16182 3	hydrolysis
5	L6	1	12 same 15
6	L7	0	hirokoshiii
7	L8	0	hirokoshii
8	L9	0	"p. hirokoshii"
9	L5	32	13 same 14
10	L10	1064	pyrococcus
11	L11	0	15 same 110
12	L12	1	12 same 110
13	L13	54539	MATSUI ISHIKAWA ISHIDA KOSUGI
14	L14	14	12 and 113



	Issue Date	Pages	Document ID	Title
1	20030626	32	US 20030119874 A1	Method for enhancing mutant enzyme activity in gaucher disease
2	20021226	27	US 20020198225 A1	Method for enhancing mutant enzyme activities in lysosomal storage disorders
3	20020801	26	US 20020102635 A1	METHODS FOR MAKING AND USING A THERMOPHILIC ENZYME AS A BETA-GLYCOSIDASE
4	20020321	25	US 20020035072 A1	Method for enhancing mutant enzyme activities in lysosomal storage disorders
5	20041116	42	US 6818803 B1	Transgenic plants as an alternative source of lignocellulosic-degrading enzymes
6	20031007	17	US 6630453 B1	Androgen derivatives and uses thereof
7	20030729	32	US 6599919 B2	Method for enhancing mutant enzyme activities in lysosomal storage disorders
8	20030708	33	US 6589964 B2	Method for enhancing mutant enzyme activities in lysosomal storage disorders
9	20030624	34	US 6583158 B1	Method for enhancing mutant enzyme activities in lysosomal storage disorders
10	20000801	92	US 6096545 A	Phosphate starvation-inducible proteins
11	19991109	20	US 5981835 A	Transgenic plants as an alternative source of lignocellulosic-degrading enzymes
12	19901030	15	US 4966856 A	Analytical element and the analytical method using the element
13	19890919	22	US 4868106 A	Analytical element and method for determining a component in a test sample

	Issue Date	Pages	Document ID	Title
14	19870728	17	US 4683198 A	Novel maltose dehydrogenase, process for its production, and analytical method using the same